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STANDAGE, SCOTT

<120> GENE CLUSTER FOR PRODUCTION OF THE ENEDIYNE ANTITUMOR  
ANTIBIOTIC C-1027

<130> 407T-896010US

<140> 09/478,188

<141> 2000-01-05

<150> 60/115,434

<151> 1999-01-06

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Met Arg Met Leu Val
1 5

acg ggc gga gcg ggt ttc atc ggc tcg cag ttc gtg cgg gcc aca ctg 163
Thr Gly Gly Ala Gly Phe Ile Gly Ser Gln Phe Val Arg Ala Thr Leu
10 15 20

cac ggc gag ctg ccg ggt tcc gag gac gcc cgg gtg acg gtc ctg gac 211
His Gly Glu Leu Pro Gly Ser Glu Asp Ala Arg Val Thr Val Leu Asp
25 30 35

aag ctg acg tac tcc ggc aat ccg gcc aac ctc acc tcc gtc gcg gcc 259
Lys Leu Thr Tyr Ser Gly Asn Pro Ala Asn Leu Thr Ser Val Ala Ala
40 45 50

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His	Pro	Arg	Tyr	Thr	Phe	Val	Gln	Gly	Asp	Thr	Val	Asp	Pro	Arg	Val	
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Val	Asp	Glu	Val	Val	Ala	Gly	His	Asp	Val	Ile	Val	His	Phe	Ala	Ala	
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Glu	Ser	His	Val	Asp	Arg	Ser	Ile	Asp	Thr	Ala	Thr	Arg	Phe	Val	Thr	
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Thr	Asn	Val	Leu	Gly	Thr	Gln	Thr	Leu	Leu	Glu	Ala	Ala	Leu	Arg	His	
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Gly	Val	Gly	Arg	Phe	Val	His	Val	Ser	Thr	Asp	Glu	Val	Tyr	Gly	Ser	
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Ile	Ala	Ser	Gly	Ser	Trp	Thr	Glu	Asp	Thr	Pro	Leu	Ala	Pro	Asn	Val	
	135					140					145					
ccc	tac	gcg	gcg	tcg	aag	gcg	ggg	tcg	gac	ctg	atg	gcg	ctc	gcc	tgg	595
Pro	Tyr	Ala	Ala	Ser	Lys	Ala	Gly	Ser	Asp	Leu	Met	Ala	Leu	Ala	Trp	
	150				155					160					165	
cac	cgc	acc	cgg	ggc	ctg	gac	gtc	gtc	gtc	acc	cgg	tgc	acc	aac	aac	643
His	Arg	Thr	Arg	Gly	Leu	Asp	Val	Val	Val	Thr	Arg	Cys	Thr	Asn	Asn	
				170					175					180		
tac	ggg	ccc	tac	cag	tac	ccc	gag	aag	gtg	atc	ccg	ctc	ttc	gtc	acc	691
Tyr	Gly	Pro	Tyr	Gln	Tyr	Pro	Glu	Lys	Val	Ile	Pro	Leu	Phe	Val	Thr	
			185					190					195			
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Asn	Ile	Leu	Asp	Gly	Leu	Arg	Val	Pro	Leu	Tyr	Gly	Asp	Gly	Ala	His	
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Arg	Arg	Asp	Trp	Leu	His	Val	Ser	Asp	His	Cys	Arg	Ala	Ile	Gln	Met	
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Val	Met	Asn	Ser	Gly	Arg	Ala	Gly	Glu	Val	Tyr	His	Ile	Gly	Gly	Gly	
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Thr	Glu	Leu	Ser	Asn	Glu	Glu	Leu	Thr	Gly	Leu	Leu	Leu	Thr	Ala	Cys	
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Gly	Thr	Asp	Trp	Ser	Cys	Val	Asp	Arg	Val	Ala	Asp	Arg	Gln	Gly	His	
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Tyr Glu Pro Leu Val Ala Phe Glu Asp Gly Leu Ala Ala Thr Val Lys	
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Trp Tyr His Glu Asn Arg Ser Trp Trp Gln Pro Leu Lys Glu Ala Ala	
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Leu Met Leu Asp Ile Asn Val Leu Met Leu Ala Leu Pro Gln Leu Ser	
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Val Val Ser Arg Ala Val Leu Gly Val Ala Gly Ala Thr Val Met Pro	
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Ser Thr Leu Ala Leu Ile Ser Asn Met Phe Glu Asp Pro Lys Glu Arg	
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Gly Thr Ala Ile Ala Met Trp Ala Ser Ala Met Met Ala Gly Val Ala	
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Leu Gly Pro Ala Val Gly Gly Leu Val Leu Ala Ala Phe Trp Trp Gly	
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Ser Val Phe Leu Ile Ala Val Pro Val Met Leu Leu Val Val Val Thr	
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Pro Pro Ser Arg Val Leu Leu Gly Gly Leu Leu Ile Ala Ala Val Gly	
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Gln Leu Leu Ile Thr Gln Val Asp Thr Glu Asp Thr Ala Leu Leu Ile	
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Ala Ala Thr Thr Leu Ile Tyr Phe Gly Ala Ser Pro Val Gly Pro Ile	
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Ile Ala Gly Leu Gly Ser Leu Gly Thr Val Val Tyr Ser Ala Gly Val	
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Glu Val Pro Asp Ala Ala Gly Pro Ala Asp Ala Asp Ala Ala Gln Glu	
765 770 775	
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Ala Asp Ala Leu Leu Asp Ser Ala Arg Ala Ala Phe Thr Ser Gly Val	
795 800 805	
cag tcc gtc gcc gcc gtc tgc gcc gtg ttc tcc ctg gcg ctc gcc gtc	2618
Gln Ser Val Ala Ala Val Cys Ala Val Phe Ser Leu Ala Leu Ala Val	
810 815 820	
ctc atc ggc acc ccg ctg ccg gac att tcc gcg atg gac cac ggg cac	2666
Leu Ile Gly Thr Arg Leu Arg Asp Ile Ser Ala Met Asp His Gly His	
825 830 835 840	
ggc gag gaa ccg gcc gag aac gac gct caa ccg gcc aca tgagcgact	2715
Gly Glu Glu Pro Ala Glu Asn Asp Ala Gln Pro Ala Thr	
845 850	

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tccggagatg caacggccgc cgtcgaggtg tgaggatcac cttccggggg gcacctgcac 2775

ggcaacggag gcgtagtgga gtactggaac agcacggcgg agaccatgcc ccgccaggaa 2835

ctcgaacagt ggaagtggcg caggctccag gccgccatgg accacgccag aaggctttcg 2895

cccttctggc gggaacgact ccccgagaac atcacctcca tggcggacta cgcggcgcg 2955

gtgcctctcc tgcgcaaggc cgacctctc gccgcggaag ccgcgtctcc cccttacggc 3015

acctggccct cgtggatcc 3035

<210> 113

<211> 332

<212> PRT

<213> Streptomyces globisporus

<220>

<223> sgA

&lt;400&gt; 113

Met	Arg	Met	Leu	Val	Thr	Gly	Gly	Ala	Gly	Phe	Ile	Gly	Ser	Gln	Phe
1				5					10					15	
Val	Arg	Ala	Thr	Leu	His	Gly	Glu	Leu	Pro	Gly	Ser	Glu	Asp	Ala	Arg
			20					25					30		
Val	Thr	Val	Leu	Asp	Lys	Leu	Thr	Tyr	Ser	Gly	Asn	Pro	Ala	Asn	Leu
		35					40					45			
Thr	Ser	Val	Ala	Ala	His	Pro	Arg	Tyr	Thr	Phe	Val	Gln	Gly	Asp	Thr
	50					55					60				
Val	Asp	Pro	Arg	Val	Val	Asp	Glu	Val	Val	Ala	Gly	His	Asp	Val	Ile
65					70					75					80
Val	His	Phe	Ala	Ala	Glu	Ser	His	Val	Asp	Arg	Ser	Ile	Asp	Thr	Ala
				85					90					95	
Thr	Arg	Phe	Val	Thr	Thr	Asn	Val	Leu	Gly	Thr	Gln	Thr	Leu	Leu	Glu
			100					105					110		
Ala	Ala	Leu	Arg	His	Gly	Val	Gly	Arg	Phe	Val	His	Val	Ser	Thr	Asp
		115					120					125			
Glu	Val	Tyr	Gly	Ser	Ile	Ala	Ser	Gly	Ser	Trp	Thr	Glu	Asp	Thr	Pro
	130					135					140				
Leu	Ala	Pro	Asn	Val	Pro	Tyr	Ala	Ala	Ser	Lys	Ala	Gly	Ser	Asp	Leu
145					150					155					160
Met	Ala	Leu	Ala	Trp	His	Arg	Thr	Arg	Gly	Leu	Asp	Val	Val	Val	Thr
				165					170					175	
Arg	Cys	Thr	Asn	Asn	Tyr	Gly	Pro	Tyr	Gln	Tyr	Pro	Glu	Lys	Val	Ile
			180					185					190		

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Pro	Leu	Phe	Val	Thr	Asn	Ile	Leu	Asp	Gly	Leu	Arg	Val	Pro	Leu	Tyr
		195					200					205			
Gly	Asp	Gly	Ala	His	Arg	Arg	Asp	Trp	Leu	His	Val	Ser	Asp	His	Cys
	210					215					220				
Arg	Ala	Ile	Gln	Met	Val	Met	Asn	Ser	Gly	Arg	Ala	Gly	Glu	Val	Tyr
225					230					235					240
His	Ile	Gly	Gly	Gly	Thr	Glu	Leu	Ser	Asn	Glu	Glu	Leu	Thr	Gly	Leu
				245					250					255	
Leu	Leu	Thr	Ala	Cys	Gly	Thr	Asp	Trp	Ser	Cys	Val	Asp	Arg	Val	Ala
			260					265					270		
Asp	Arg	Gln	Gly	His	Asp	Arg	Arg	Tyr	Ser	Leu	Asp	Ile	Thr	Lys	Ile
		275					280					285			
Arg	Gln	Glu	Leu	Gly	Tyr	Glu	Pro	Leu	Val	Ala	Phe	Glu	Asp	Gly	Leu
	290					295					300				

Ala Ala Thr Val Lys Trp Tyr His Glu Asn Arg Ser Trp Trp Gln Pro  
 305 310 315 320

Leu Lys Glu Ala Ala Gly Leu Leu Asp Ala Val Gly  
 325 330

<210> 114

<211> 521

<212> PRT

<213> Streptomyces globisporus

<220>

<223> sgcB

<400> 114

Met Thr Ala Val Lys Glu Pro Thr Ser Arg Ala Gly Arg Arg Glu Trp  
 1 5 10 15

Ile Ala Leu Val Val Leu Ser Leu Pro Thr Met Leu Leu Met Leu Asp  
 20 25 30

Ile Asn Val Leu Met Leu Ala Leu Pro Gln Leu Ser Glu Asp Leu Gly  
 35 40 45

Ala Ser Ser Thr Gln Gln Leu Trp Ile Thr Asp Ile Tyr Gly Phe Ala  
 50 55 60

Ile Ala Gly Phe Leu Val Thr Met Gly Thr Leu Gly Asp Arg Ile Gly  
 65 70 75 80

Arg Arg Arg Leu Leu Leu Gly Gly Ala Ala Val Phe Ala Val Val Ser  
 85 90 95

Val Val Ala Ala Phe Ser Asp Ser Ala Ala Met Leu Val Val Ser Arg  
 100 105 110

Ala Val Leu Gly Val Ala Gly Ala Thr Val Met Pro Ser Thr Leu Ala  
 115 120 125

Leu Ile Ser Asn Met Phe Glu Asp Pro Lys Glu Arg Gly Thr Ala Ile  
 130 135 140

Ala Met Trp Ala Ser Ala Met Met Ala Gly Val Ala Leu Gly Pro Ala  
 145 150 155 160

Val Gly Gly Leu Val Leu Ala Ala Phe Trp Trp Gly Ser Val Phe Leu  
 165 170 175

Ile Ala Val Pro Val Met Leu Leu Val Val Val Thr Gly Pro Val Leu  
 180 185 190

Leu Thr Glu Ser Arg Asp Pro Asp Ala Gly Arg Leu Asp Leu Leu Ser  
 195 200 205

Ala Gly Leu Ser Leu Ala Thr Val Leu Pro Val Ile Tyr Gly Leu Lys  
 210 215 220

Glu Leu Ala Arg Thr Gly Trp Asp Pro Leu Ala Ala Gly Ala Val Val  
 225 230 235 240  
 Leu Gly Val Ile Phe Gly Ala Leu Phe Val Gln Arg Gln Arg Arg Leu  
 245 250 255  
 Ala Asp Pro Met Leu Asp Leu Gly Leu Phe Ala Asp Arg Thr Leu Arg  
 260 265 270  
 Ala Gly Leu Thr Val Ser Leu Val Asn Ala Val Ile Met Gly Gly Thr  
 275 280 285  
 Gly Leu Met Val Ala Leu Tyr Leu Gln Thr Ile Ala Gly His Ser Pro  
 290 295 300  
 Leu Ala Ala Gly Leu Trp Leu Leu Ile Pro Ala Cys Met Leu Val Val  
 305 310 315 320  
 Gly Val Gln Leu Ser Asn Leu Leu Ala Gln Arg Met Pro Pro Ser Arg  
 325 330 335  
 Val Leu Leu Gly Gly Leu Leu Ile Ala Ala Val Gly Gln Leu Leu Ile  
 340 345 350  
 Thr Gln Val Asp Thr Glu Asp Thr Ala Leu Leu Ile Ala Ala Thr Thr  
 355 360 365  
 Leu Ile Tyr Phe Gly Ala Ser Pro Val Gly Pro Ile Thr Thr Gly Ala  
 370 375 380  
 Ile Met Gly Ala Ala Pro Pro Glu Lys Ala Gly Ala Ala Ser Ser Leu  
 385 390 395 400  
 Ser Ala Thr Gly Gly Glu Phe Gly Val Ala Leu Gly Ile Ala Gly Leu  
 405 410 415  
 Gly Ser Leu Gly Thr Val Val Tyr Ser Ala Gly Val Glu Val Pro Asp  
 420 425 430  
 Ala Ala Gly Pro Ala Asp Ala Asp Ala Ala Gln Glu Ser Ile Ala Gly  
 435 440 445  
 Ala Leu His Thr Ala Gly Gln Leu Ala Pro Gly Ser Ala Asp Ala Leu  
 450 455 460  
 Leu Asp Ser Ala Arg Ala Ala Phe Thr Ser Gly Val Gln Ser Val Ala  
 465 470 475 480  
 Ala Val Cys Ala Val Phe Ser Leu Ala Leu Ala Val Leu Ile Gly Thr  
 485 490 495  
 Arg Leu Arg Asp Ile Ser Ala Met Asp His Gly His Gly Glu Glu Pro  
 500 505 510  
 Ala Glu Asn Asp Ala Gln Pro Ala Thr  
 515 520

full  
 108  
 cont

&lt;210&gt; 115

&lt;211&gt; 329

&lt;212&gt; PRT

&lt;213&gt; Saccharopolyspora erythraea

&lt;400&gt; 115

Met Arg Val Leu Val Thr Gly Gly Ala Gly Phe Ile Gly Ser His Tyr  
1 5 10 15

Val Arg Gln Leu Leu Gly Gly Ala Tyr Pro Ala Phe Ala Gly Ala Asp  
20 25 30

Val Val Val Leu Asp Lys Leu Thr Tyr Ala Gly Asn Glu Glu Asn Leu  
35 40 45

Arg Pro Val Ala Asp Asp Pro Arg Phe Arg Phe Val Arg Gly Asp Ile  
50 55 60

Cys Glu Trp Asp Val Val Ser Glu Val Met Arg Glu Val Asp Val Val  
65 70 75 80

Val His Phe Ala Ala Glu Thr His Val Asp Arg Ser Ile Leu Gly Ala  
85 90 95

Ser Asp Phe Val Val Thr Asn Val Val Gly Thr Asn Thr Leu Leu Gln  
100 105 110

Gly Ala Leu Ala Ala Asn Val Ser Lys Phe Val His Val Ser Thr Asp  
115 120 125

Glu Val Tyr Gly Thr Ile Glu His Gly Ser Trp Pro Glu Asp His Leu  
130 135 140

Leu Glu Pro Asn Ser Pro Tyr Ser Ala Ala Lys Ala Gly Ser Asp Leu  
145 150 155 160

Ile Ala Arg Ala Tyr His Arg Thr His Gly Leu Pro Val Cys Ile Thr  
165 170 175

Arg Cys Ser Asn Asn Tyr Gly Pro Tyr Gln Phe Pro Glu Lys Val Leu  
180 185 190

Pro Leu Phe Ile Thr Asn Leu Met Asp Gly Arg Arg Val Pro Leu Tyr  
195 200 205

Gly Asp Gly Leu Asn Val Arg Asp Trp Leu His Val Thr Asp His Cys  
210 215 220

Arg Gly Ile Gln Leu Val Ala Glu Ser Gly Arg Ala Gly Glu Ile Tyr  
225 230 235 240

Asn Ile Gly Gly Gly Thr Glu Leu Thr Asn Lys Glu Leu Thr Glu Arg  
245 250 255

Val Leu Glu Leu Met Gly Gln Asp Trp Ser Met Val Gln Pro Val Thr  
260 265 270

Full  
DS  
Gene

Asp Arg Lys Gly His Asp Arg Arg Tyr Ser Val Asp His Thr Lys Ile  
275 280 285

Ser Glu Glu Leu Gly Tyr Glu Pro Val Val Pro Phe Glu Arg Gly Leu  
290 295 300

Ala Glu Thr Ile Glu Trp Tyr Arg Asp Asn Arg Ala Trp Trp Glu Pro  
305 310 315 320

Leu Lys Ser Ala Pro Asp Gly Gly Lys  
325

<210> 116

<211> 333

<212> PRT

<213> Streptomyces fradiae

<400> 116

Met Arg Val Leu Val Thr Gly Gly Ala Gly Phe Ile Gly Ser His Phe  
1 5 10 15

Thr Gly Gln Leu Leu Thr Gly Ala Tyr Pro Asp Leu Gly Ala Thr Arg  
20 25 30

Thr Val Val Leu Asp Lys Leu Thr Tyr Ala Gly Asn Pro Ala Asn Leu  
35 40 45

Glu His Val Ala Gly His Pro Asp Leu Glu Phe Val Arg Gly Asp Ile  
50 55 60

Ala Asp His Gly Trp Trp Arg Arg Leu Met Glu Gly Val Gly Leu Val  
65 70 75 80

Val His Phe Ala Ala Glu Ser His Val Asp Arg Ser Ile Glu Ser Ser  
85 90 95

Glu Ala Phe Val Arg Thr Asn Val Glu Gly Thr Arg Val Leu Leu Gln  
100 105 110

Ala Ala Val Asp Ala Gly Val Gly Arg Phe Val His Ile Ser Thr Asp  
115 120 125

Glu Val Tyr Gly Ser Ile Ala Glu Gly Ser Trp Pro Glu Asp His Pro  
130 135 140

Val Ala Pro Asn Ser Pro Tyr Ala Ala Thr Lys Lys Ala Ser Asp Leu  
145 150 155 160

Leu Ala Leu Ala Tyr His Arg Thr Tyr Gly Leu Asp Val Arg Val Thr  
165 170 175

Arg Cys Ser Asn Asn Tyr Gly Pro Arg Gln Tyr Pro Glu Lys Ala Val  
180 185 190

Pro Leu Phe Thr Thr Asn Leu Leu Asp Gly Leu Pro Val Pro Leu Tyr  
195 200 205

Shu  
DB  
CMT

Gly Asp Gly Gly Asn Thr Arg Glu Trp Leu His Val Asp Asp His Cys  
 210 215 220  
 Arg Gly Val Ala Leu Val Gly Ala Gly Gly Arg Pro Gly Val Ile Tyr  
 225 230 235 240  
 Asn Ile Gly Gly Gly Thr Glu Leu Thr Asn Ala Glu Leu Thr Asp Arg  
 245 250 255  
 Ile Leu Glu Leu Cys Gly Ala Asp Arg Ser Ala Leu Arg Arg Val Ala  
 260 265 270  
 Asp Arg Pro Gly His Asp Arg Arg Tyr Ser Val Asp Thr Thr Lys Ile  
 275 280 285  
 Arg Glu Glu Leu Gly Tyr Ala Pro Arg Thr Gly Ile Thr Glu Gly Leu  
 290 295 300  
 Ala Gly Thr Val Ala Trp Tyr Arg Asp Asn Arg Ala Trp Trp Glu Pro  
 305 310 315 320  
 Leu Lys Arg Ser Pro Gly Gly Arg Glu Leu Glu Arg Ala  
 325 330

<210> 117  
 <211> 331  
 <212> PRT  
 <213> Streptomyces argillaceus

<400> 117  
 Met Thr Thr Thr Ser Ile Leu Val Thr Gly Gly Ala Gly Phe Ile Gly  
 1 5 10 15  
 Ser His Tyr Val Arg Thr Leu Leu Gly Pro Arg Gly Val Pro Asp Val  
 20 25 30

Thr Val Thr Val Leu Asp Lys Leu Thr Tyr Ala Gly Thr Leu Thr Asn  
 35 40 45  
 Leu Ala Glu Val Ser Asp Ser Asp Arg Phe Arg Phe Val Arg Gly Asp  
 50 55 60  
 Ile Cys Asp Ala Pro Leu Val Asp Asp Leu Leu Ala Val His Asp Gln  
 65 70 75 80  
 Val Val His Phe Ala Ala Glu Ser His Val Asp Arg Ser Ile Leu Gly  
 85 90 95  
 Ala Ala Asp Phe Val Arg Thr Asn Val Thr Gly Thr Gln Thr Leu Leu  
 100 105 110  
 Asp Ala Ala Leu Arg Gln Gly Ile Glu Thr Phe Val His Ile Ser Thr  
 115 120 125  
 Asp Glu Val Tyr Gly Ser Ile Asp Ala Gly Ser Trp Pro Glu Thr Ala  
 130 135 140

See  
 D8  
 cont

Pro Val Ser Pro Asn Ser Leu Tyr Ser Ala Ala Lys Ala Ser Ser Asp  
 145 150 155 160  
 Leu Val Ala Leu Ala Tyr His Arg Thr His Gly Leu Asp Val Arg Val  
 165 170 175  
 Thr Arg Cys Ser Asn Asn Tyr Gly Ser His Gln Phe Pro Glu Lys Val  
 180 185 190  
 Ile Pro Leu Phe Val Thr Ser Leu Leu Asp Gly Arg Glu Val Pro Leu  
 195 200 205  
 Tyr Gly Asp Gly Thr Asn Val Arg Asp Trp Leu His Val Asp Asp His  
 210 215 220  
 Val Arg Ala Ile Glu Leu Val Arg Thr Gly Gly Arg Ala Gly Glu Val  
 225 230 235 240  
 Tyr Asn Ile Gly Gly Gly Thr Glu Leu Ser Asn Lys Glu Leu Thr Gln  
 245 250 255  
 Leu Leu Leu Asp Ala Cys Gly Ala Gly Trp Asp Arg Val Arg Tyr Val  
 260 265 270  
 Thr Asp Arg Lys Gly His Asp Arg Arg Tyr Ser Val Asp Cys Thr Lys  
 275 280 285  
 Ile Arg Arg Glu Leu Gly Tyr Arg Pro Ala Arg Glu Phe Gly Asp Ala  
 290 295 300  
 Leu Ala Glu Thr Val Ala Trp Tyr Arg His His Arg Ala Trp Trp Glu  
 305 310 315 320  
 Pro Leu Thr Arg Ala Tyr Gly Ala Val Ala Ala  
 325 330

&lt;210&gt; 118

&lt;211&gt; 6

&lt;212&gt; PRT

&lt;213&gt; Artificial Sequence

&lt;220&gt;

&lt;223&gt; Description of Artificial Sequence: 6-His tag

&lt;400&gt; 118

His His His His His His

1

5

See  
 D8  
 cont